



February 9, 2017

Food and Drug Administration
10903 New Hampshire Avenue
Document Control Center – WO66-G609
Silver Spring, MD 20993-0002

CEPHEID
SCOTT A. CAMPBELL
CORPORATE VICE PRESIDENT & CHIEF REGULATORY OFFICER
904 CARIBBEAN DRIVE
SUNNYVALE, CA 94089

Re: K162331

Trade/Device Name: Xpert[®] Xpress Flu/RSV
Regulation Number: 21 CFR 866.3980
Regulation Name: Respiratory viral panel multiplex nucleic acid assay
Regulatory Class: II
Product Code: OCC, OOI, JSM
Dated: August, 18 2016
Received: August 23, 2016

Dear Dr. Campbell:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820); and if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

If you desire specific advice for your device on our labeling regulations (21 CFR Parts 801 and 809), please contact the Division of Industry and Consumer Education at its toll-free number (800) 638 2041 or (301) 796-7100 or at its Internet address <http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm>. Also, please note the regulation entitled, “Misbranding by reference to premarket notification” (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to <http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm> for the CDRH’s Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Industry and Consumer Education at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address <http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm>.

Sincerely yours,


Steven R. Gitterman -S

Uwe Scherf, M.Sc., Ph.D.
Director
Division of Microbiology Devices
Office of In Vitro Diagnostics
and Radiological Health
Center for Devices and Radiological Health

Enclosure

Indications for Use

510(k) Number (if known)

K162331

Device Name

Xpert Xpress Flu/RSV

Indications for Use (Describe)

The Cepheid Xpert® Xpress Flu/RSV Assay, performed on the GeneXpert® Instrument Systems, is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the *in vitro* qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Xpress Flu/ RSV Assay uses nasopharyngeal (NP) swab specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Xpress Flu/RSV Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus infections in conjunction with clinical and epidemiological risk factors.

Negative results do not preclude influenza virus or RSV infection and should not be used as the sole basis for treatment or other patient management decisions.

Performance characteristics for influenza A were established during the 2015-2016 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.

If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health departments for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.

Ancillary Collection Kit Indications for Use:

The Xpert® Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay or the Xpert Flu/RSV XC Assay.

The Xpert® Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay or the Xpert Xpress Flu/RSV Assay.

Type of Use (Select one or both, as applicable)

Prescription Use (Part 21 CFR 801 Subpart D)

Over-The-Counter Use (21 CFR 801 Subpart C)

PLEASE DO NOT WRITE BELOW THIS LINE – CONTINUE ON A SEPARATE PAGE IF NEEDED.

FOR FDA USE ONLY

Concurrence of Center for Devices and Radiological Health (CDRH) (Signature)

This section applies only to requirements of the Paperwork Reduction Act of 1995.

DO NOT SEND YOUR COMPLETED FORM TO THE PRA STAFF EMAIL ADDRESS BELOW.

The burden time for this collection of information is estimated to average 79 hours per response, including the time to review instructions, search existing data sources, gather and maintain the data needed and complete and review the collection of information. Send comments regarding this burden estimate or any other aspect of this information collection, including suggestions for reducing this burden, to:

Department of Health and Human Services
Food and Drug Administration
Office of Chief Information Officer
Paperwork Reduction Act (PRA) Staff
PRAStaff@fda.hhs.gov

“An agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a currently valid OMB number.”

CONFIDENTIAL

510(k) Summary

As required by 21 CFR Section 807.92(c).

Submitted by: Cepheid
904 Caribbean Drive
Sunnyvale, CA 90489
Phone number: (847) 228-3299
Fax number: (847) 890-6589

Contact: Scott A. Campbell, PhD, MBA

Date of Preparation: August 18, 2016

Device:

Trade name: Xpert[®] Xpress Flu/RSV

Common name: Xpert Xpress Flu/RSV Assay

Type of Test: Automated, multiplex real-time reverse transcription-polymerase chain reaction (RT-PCR) assay intended for the *in vitro* qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA.

Regulation number/ Classification name/ Product code: 866.3980/Respiratory viral panel multiplex nucleic acid assay/OCC

866.2570/Instrumentation for clinical multiplex test systems/OOI

866.2390/Culture Media, Non-propagating Transport/JSM

Classification Advisory Panel: Class II
Microbiology (83)

Prescription Use: Yes

Predicate Devices Assay: 1) *For the detection and differentiation of influenza A, influenza B, and RSV A/B viral RNA in nasopharyngeal swab specimens:*
Xpert[®] Flu/RSV XC Assay [510(k) #K142045]

2) *For the Sample Collection Kits:*

Cepheid Xpert Nasopharyngeal Sample Collection Kit [510(k) # K151226]

CONFIDENTIAL

Device Description:

The Xpert Xpress Flu/RSV Assay is a rapid, automated *in vitro* diagnostic test for qualitative detection and differentiation of influenza A (Flu A), influenza B (Flu B), and respiratory syncytial virus (RSV) viral RNA directly from nasopharyngeal (NP) swab specimens. The assay is performed on the Cepheid GeneXpert[®] Instrument Systems.

The Xpert Xpress Flu/RSV Assay includes reagents for the simultaneous detection and differentiation of the target viruses. The primers and probes in the Xpert Xpress Flu/RSV Assay detect the presence of nucleic acid sequences for Flu A, Flu B, and RSV directly from NP swab specimens collected from patients with signs and symptoms of respiratory infection. A Sample Processing Control (SPC) and a Probe Check Control (PCC) are internal controls utilized by the GeneXpert Instrument System platform. The SPC is present in every assay to control for adequate processing of the target viruses and to monitor for the presence of inhibitor(s) in the PCR assay to avoid false-negative results. The PCC verifies reagent rehydration, real-time PCR tube filling in the cartridge, probe integrity, and dye stability.

The specimens are collected in viral transport medium and transported to the GeneXpert area. The specimen is prepared according to package insert instructions and transferred to the sample chamber (large opening) of the Xpert Xpress Flu/RSV Assay cartridge. The GeneXpert cartridge is loaded onto the GeneXpert Instrument System platform, which performs hands-off automated sample processing and real-time RT-PCR for detection of Flu and RSV viral RNA. Summary and detailed test results are obtained in approximately 30 minutes or less. The results are automatically generated at the end of the process in a report that can be viewed and printed.

CONFIDENTIAL

Device Intended Use:

The Cepheid Xpert[®] Xpress Flu/RSV Assay, performed on the GeneXpert[®] Instrument Systems, is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the *in vitro* qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Xpress Flu/RSV Assay uses nasopharyngeal (NP) swab specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Xpress Flu/RSV Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus infections in conjunction with clinical and epidemiological risk factors.

Negative results do not preclude influenza virus or RSV infection and should not be used as the sole basis for treatment or other patient management decisions.

Performance characteristics for influenza A were established during the 2015-2016 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.

If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health departments for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.

Ancillary Specimen Collection Kit

The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of

CONFIDENTIAL

respiratory infection prior to analysis with the Xpert Flu Assay or the Xpert Flu/RSV XC Assay.

The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay or the Xpert Xpress Flu/RSV Assay.

Substantial Equivalence:

The Xpert Xpress Flu/RSV Assay is substantially equivalent to the current Xpert[®] Flu/RSV XC Assay [510(k) #K142045]. The Xpert Xpress Flu/RSV Assay detects influenza A, influenza B, and RSV A/B from nasopharyngeal (NP) swab specimens and the Xpert[®] Flu/RSV XC Assay detects influenza A, influenza B, and RSV from both NP swab specimens and nasal aspirates/washes (NA/W) specimens. Both assays utilize the same technology by determining the presence of the target organisms through real-time RT-PCR amplification and fluorogenic target-specific hybridization detection. A multi-center clinical study was conducted to determine the performance characteristics of the Xpert Xpress Flu/RSV Assay relative to the predicate device, which has been FDA cleared for NP swab and NA/W specimens. Discordant results between the Xpert Xpress Flu/RSV Assay and the reference method Prodesse ProFlu+ Assay [510(k) # K132129] were analyzed by bi-directional sequencing using primers different from those used in the Xpert Xpress Flu/RSV Assay. The study results showed that the Xpert Xpress Flu/RSV Assay is substantially equivalent to the predicate device.

Table 8-1 shows the similarities and differences between the Xpert Xpress Flu/RSV Assay and the predicate devices.

CONFIDENTIAL

Table 8-1: Comparison of Similarities and Differences of the Xpert Xpress Flu/RSV Assay with the Predicate Devices

Similarities		
	Device	Predicate
Item	Cepheid Xpert [®] Xpress Flu/RSV	Cepheid Xpert [®] Flu/RSV XC 510(k)# K142045
Regulation	866.3980	Same
Product Code	OCC, OOI	Same
Device Class	II	Same
Technology Principle of Operation	Multiplex real time RT-PCR	Same
Intended Use	<p>The Cepheid Xpert[®] Xpress Flu/RSV Assay, performed on the GeneXpert Instrument Systems, is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the <i>in vitro</i> qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Xpress Flu/RSV Assay uses nasopharyngeal (NP) swab specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Xpress Flu/RSV Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus infections in conjunction with clinical and epidemiological risk factors. Negative results do not</p>	<p>The Cepheid Xpert[®] Flu/RSV XC Assay is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the <i>in vitro</i> qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Flu/RSV XC Assay uses nasopharyngeal swab and nasal aspirate/wash specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Flu/RSV XC Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus infections in conjunction with clinical and epidemiological risk factors. Negative results do not</p>

CONFIDENTIAL

Similarities		
	Device	Predicate
Item	Cepheid Xpert® Xpress Flu/RSV	Cepheid Xpert® Flu/RSV XC 510(k)# K142045
	<p>preclude influenza virus or RSV infection and should not be used as the sole basis for treatment or other patient management decisions.</p> <p>Performance characteristics for influenza A were established during the 2015-2016 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.</p> <p>If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health departments for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.</p>	<p>preclude influenza virus or respiratory syncytial virus infection and should not be used as the sole basis for treatment or other patient management decisions.</p> <p>Performance characteristics for influenza A were established during the 2013-2014 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.</p> <p>If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health departments for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.</p>

CONFIDENTIAL

Similarities		
	Device	Predicate
Item	Cepheid Xpert[®] Xpress Flu/RSV	Cepheid Xpert[®] Flu/RSV XC 510(k)# K142045
Indications for Use	Patients with signs and symptoms of respiratory infection in conjunction with clinical and epidemiological risk factors	Same
Assay Targets	Influenza A Virus, Influenza B Virus, and RSV viral RNA	Same
Nucleic Acid Extraction	Yes	Same
Extraction Methods	Sample preparation integrated in GeneXpert Cartridge and GeneXpert Instrumentation System	Same
Assay Results	Qualitative	Same
Instrument System	Cepheid GeneXpert Instrument Systems; same Cepheid I-core technology	Same
Assay Controls	Encapsulated (armored) RNA pseudovirus as a sample processing control. Available but not provided are inactivated virus controls for influenza A/B and RSV as external positive controls, and Coxsackie virus as an external negative control.	Same
Primers and probes	Primers and probes to detect the presence of nucleic acid sequences of influenza A, influenza B, and RSV.	Primers and probes to detect the presence of influenza A, influenza A subtype H7N9, influenza B and RSV.
Laboratory Users	Laboratory users in moderate and high complexity laboratory settings.	Same

CONFIDENTIAL

Similarities		
	Device	Predicate
Item	Cepheid Xpert® Xpress Flu/RSV	Cepheid Xpert® Flu/RSV XC 510(k)# K142045
Sample Preparation	Self-contained and automated after mixed specimen is added to cartridge. All other reagents are contained in the cartridge.	Same
Primers and probes for influenza A, influenza B, and influenza A subtype H1N1	Primers and probes to detect the presence of nucleic acid sequences of influenza A, influenza B, and RSV A/B. The Xpert Xpress Flu/RSV Assay contains primers and probes to detect additional RNA segments in order to protect the assay sensitivity and specificity from mutations in the influenza genome due to antigenic drifts and shifts.	Primers and probes to detect the presence of nucleic acid sequences of influenza A, influenza B, and RSV A/B. The Xpert Flu/RSV XC Assay contains primers and probes to detect additional RNA segments in order to protect the assay sensitivity and specificity from mutations in the influenza genome due to antigenic drifts and shifts.
Target Sequence	Influenza A: Matrix protein (MP), basic polymerase (PB2) and acidic protein (PA) Influenza B: Matrix protein (MP) and Non-structural proteins (NS 1 and NS 2)	Same
	RSV A and RSV B: Nucleocapsid protein	Same
Internal Controls	Sample processing control (SPC) and probe check control (PCC).	Same
Combinatorial Assay Selections	Yes, user may select combined assay with all targets or a Flu only assay or a RSV only assay.	Same
Early assay termination function	On Flu only or RSV only assay selections	Same

CONFIDENTIAL

Differences		
Item	Device	Predicate
	Cepheid Xpert Flu/RSV XC	Cepheid Xpert Flu/RSV XC
Specimen Types	Nasopharyngeal (NP) swab specimens	Nasal aspirate/wash (NA/W) specimens and Nasopharyngeal (NP) swab specimens
Time to obtain test results	Approximately 30 minutes or less for sample preparation and RT-PCR	Approximately 60 minutes or less for sample preparation and RT-PCR

The Xpert Xpress Flu/RSV Assay has the same intended use and technological characteristics as the predicate device. The clinical study demonstrates that the Xpert Xpress Flu/RSV Assay is substantially equivalent to the predicate device.

The predicate device for the ancillary specimen collection kit, the Xpert[®] Nasopharyngeal Sample Collection Kit is the Cepheid Nasopharyngeal Sample Collection Kit, [510(k) # K151226]. The similarities and differences are shown in Table 8-2.

CONFIDENTIAL

Table 8-2: Comparison of Similarities and Differences of the Xpert Nasopharyngeal Sample Collection Kit with the Predicate Device

Similarities		
Item	Device	Predicate
	Xpert [®] Nasopharyngeal Sample Collection Kit	Xpert [®] Nasopharyngeal Sample Collection Kit 510(k)# K151226
Intended Use	<p>The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay or the Xpert Flu/RSV XC Assay. The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay and the Xpert Xpress Flu/RSV Assay.</p> <p>The Xpert Nasopharyngeal Sample Collection Kit has been cleared for use only with the Xpert Flu Assay, Xpert Flu/RSV XC Assay, Xpert Flu+RSV Xpress Assay and Xpert Xpress Flu/RSV Assay.</p>	<p>The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay or the Xpert Flu/RSV XC Assay. The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay.</p> <p>The Xpert Nasopharyngeal Sample Collection Kit has been cleared for use only with the Xpert Flu Assay, Xpert Flu/RSV XC Assay and Xpert Flu+RSV Xpress Assay.</p>
Single-use Device	Yes	Same

CONFIDENTIAL

Similarities		
Item	Device	Predicate
	Xpert[®] Nasopharyngeal Sample Collection Kit	Xpert[®] Nasopharyngeal Sample Collection Kit 510(k)# K151226
Transport Medium Formulation	Hank's Balanced Salt Solution Bovine Serum Albumin L-cysteine Gelatin Sucrose L-glutamic acid HEPES buffer Vancomycin Amphotericin B Colistin Phenol red	Same
pH	7.3 ± 0.2	Same
Storage Temperature	2 - 25°C (refrigerated and room temperature)	Same
Volume	3 ml	Same
Glass Beads	3 x 3 mm	Same
Container	Plastic (medical-grade polypropylene)	Same
Product Configuration	Medium Tube in Kit with individually-wrapped sterile swab.	Same

CONFIDENTIAL

Differences		
Item	Device	Predicate
	Xpert Nasopharyngeal Sample Collection Kit	Xpert Nasopharyngeal Sample Collection Kit 510(k)# K151226
Intended Use	For collection, preservation and transport of nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay, Xpert Flu/RSV XC Assay, Xpert Flu+RSV Xpress Assay and Xpert Xpress Flu/RSV Assay.	For collection, preservation and transport of nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay, Xpert Flu/RSV XC Assay and Xpert Flu+RSV Xpress Assay.

Both devices have the same general intended use and use the same technology to collect, store and transport clinical specimens, including viruses, to the laboratory for further testing. The prospective component of the multi-center clinical study of the Xpert Xpress Flu/RSV Assay was conducted using Xpert Nasopharyngeal Sample Collection Kit [510(k) # K151226] demonstrating that the Xpert Nasopharyngeal Sample Collection Kit is substantially equivalent to the predicate device.

Non-Clinical Studies:

Analytical Sensitivity (Limit of Detection)

Studies were performed to determine the analytical limit of detection (LoD) of the Xpert Xpress Flu/RSV Assay with two lots of reagents across three testing days. The higher LoD observed per strain and per lot as determined by probit analysis was selected for verification. Verification of the estimated LoD claim was performed on one reagent lot across a minimum of three

CONFIDENTIAL

testing days. LoD was established using two influenza A H3N2 strains, two influenza A 2009 H1N1 strains, two influenza B strains, two respiratory syncytial virus A (RSV A) strains and two respiratory syncytial virus B (RSV B) strains. Viruses were diluted into negative pooled NP swab clinical matrix for testing. The LoD is defined as the lowest concentration (tissue culture infective dose, TCID₅₀/mL) per sample that can be reproducibly distinguished from negative samples with 95% confidence or the lowest concentration at which 19 of 20 replicates were positive. Each strain was tested in replicates of 20 per concentration of virus. The LoD values for each strain tested are summarized in Tables 8-3 – 8-7.

**Table 8-3 Confirmed LoD (TCID₅₀/mL):
Influenza A 2009 H1N1**

Virus Strain	Confirmed LoD (TCID₅₀/mL)
Influenza A/California/7/2009	0.02
Influenza A/Florida/27/2011	0.04

Table 8-4 Confirmed LoD (TCID₅₀/mL): Influenza A H3N2

Virus Strain	Confirmed LoD (TCID₅₀/mL)
Influenza A/Perth/16/2009	0.01
Influenza A/Victoria/361/2011	0.75

Table 8-5 Confirmed LoD (TCID₅₀/mL): Influenza B

Virus Strain	Confirmed LoD (TCID₅₀/mL)
Influenza B/Mass/2/2012	0.40
Influenza B/Wisconsin/01/2011	0.19

CONFIDENTIAL

Table 8-6 Confirmed LoD (TCID₅₀/mL) Respiratory Syncytial Virus A

Virus Strain	Confirmed LoD (TCID₅₀/mL)
RSV A/2/Australia/61	0.87
RSV A/Long/MD/56	1.10

Table 8-7 Confirmed LoD (TCID₅₀/mL): Respiratory Syncytial Virus B

Virus Strain	Confirmed LoD (TCID₅₀/mL)
RSV B/Wash/18537/62	0.79
RSV B/9320/MA/77	2.30

Analytical Specificity (Exclusivity)

The analytical specificity of the Xpert Xpress Flu/RSV Assay was evaluated by testing a panel of 44 cultures consisting of 16 viral, 26 bacterial, and two yeast strains representing common respiratory pathogens or those potentially encountered in the nasopharynx. Three replicates of each bacterial and yeast strain were tested at concentrations of $\geq 1 \times 10^6$ CFU/mL with the exception of one strain that was tested at 1×10^5 CFU/mL (*Chlamydia pneumoniae*). Three replicates of each virus were tested at concentrations of $\geq 1 \times 10^5$ TCID₅₀/mL. The analytical specificity was 100%. Results are shown in Table 8-8.

CONFIDENTIAL

Table 8-8 Analytical Specificity of the Xpert Xpress Flu/RSV Assay

Organism	Concentration	Result		
		Influenza A	Influenza B	RSV
No Template Control	N/A	NEG	NEG	NEG
Adenovirus Type 1	1.12E+06 TCID ₅₀ /mL	NEG	NEG	NEG
Adenovirus Type 7	1.87E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Human coronavirus OC43	2.85E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Human coronavirus 229E	1.00E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Cytomegalovirus	1.00E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Echovirus	3.31E+07 TCID ₅₀ /mL	NEG	NEG	NEG
Enterovirus	3.55E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Epstein Barr Virus	7.16E+07 TCID ₅₀ /mL	NEG	NEG	NEG
HSV	8.90E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Measles	6.31E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Human metapneumovirus	1.00E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Mumps virus	6.31E+06 TCID ₅₀ /mL	NEG	NEG	NEG
Human parainfluenza Type 1	1.15E+06 TCID ₅₀ /mL	NEG	NEG	NEG
Human parainfluenza Type 2	6.31E+05 TCID ₅₀ /mL	NEG	NEG	NEG
Human parainfluenza Type 3	3.55E+06 TCID ₅₀ /mL	NEG	NEG	NEG
Rhinovirus Type 1A	1.26E+05 TCID ₅₀ /mL	NEG	NEG	NEG
<i>Acinetobacter baumannii</i>	1.00E+06 CFU/mL	NEG	NEG	NEG
<i>Burkholderia cepacia</i>	3.30E+06 CFU/mL	NEG	NEG	NEG
<i>Candida albicans</i>	3.20E+06 CFU/mL	NEG	NEG	NEG
<i>Candida parapsilosis</i>	3.00E+06 CFU/mL	NEG	NEG	NEG
<i>Bordetella pertussis</i>	3.30E+06 CFU/mL	NEG	NEG	NEG
<i>Chlamydia pneumoniae</i>	1.00E+05 CFU/mL	NEG	NEG	NEG
<i>Citrobacter freundii</i>	3.30E+06 CFU/mL	NEG	NEG	NEG
<i>Corynebacterium sp.</i>	3.30E+06 CFU/mL	NEG	NEG	NEG
<i>Escherichia coli</i>	1.00E+07 CFU/mL	NEG	NEG	NEG
<i>Enterococcus faecalis</i>	1.30E+06 CFU/mL	NEG	NEG	NEG
<i>Haemophilus influenzae</i>	1.00E+06 CFU/mL	NEG	NEG	NEG
<i>Lactobacillus reuteri</i>	1.00E+06 CFU/mL	NEG	NEG	NEG
<i>Legionella spp.</i>	1.00E+06 CFU/mL	NEG	NEG	NEG
<i>Moraxella catarrhalis</i>	1.00E+07 CFU/mL	NEG	NEG	NEG
<i>Mycobacterium tuberculosis</i> (avirulent)	1.00E+06 CFU/mL	NEG	NEG	NEG
<i>Mycoplasma pneumoniae</i>	1.00E+06 CFU/mL	NEG	NEG	NEG

CONFIDENTIAL

Organism	Concentration	Result		
		Influenza A	Influenza B	RSV
<i>Neisseria meningitidis</i>	2.15E+06 CFU/mL	NEG	NEG	NEG
<i>Neisseria mucosa</i>	1.00E+07 CFU/mL	NEG	NEG	NEG
<i>Propionibacterium acnes</i>	2.40E+07 CFU/mL	NEG	NEG	NEG
<i>Pseudomonas aeruginosa</i>	3.70E+06 CFU/mL	NEG	NEG	NEG
<i>Staphylococcus aureus</i> (protein A producer)	2.20E+06 CFU/mL	NEG	NEG	NEG
<i>Staphylococcus epidermidis</i>	3.40E+06 CFU/mL	NEG	NEG	NEG
<i>Staphylococcus haemolyticus</i>	4.00E+06 CFU/mL	NEG	NEG	NEG
<i>Streptococcus agalactiae</i>	3.50E+06 CFU/mL	NEG	NEG	NEG
<i>Streptococcus pneumoniae</i>	1.00E+06 CFU/mL	NEG	NEG	NEG
<i>Streptococcus pyogenes</i>	1.00E+07 CFU/mL	NEG	NEG	NEG
<i>Streptococcus salivarius</i>	1.00E+07 CFU/mL	NEG	NEG	NEG
<i>Streptococcus sanguinis</i>	3.10E+06 CFU/mL	NEG	NEG	NEG

Analytical Reactivity (Inclusivity)

The analytical reactivity of the Xpert Xpress Flu/RSV Assay was evaluated against multiple strains of influenza A H1N1 (seasonal pre-2009), influenza A H1N1 (pandemic 2009), influenza A H3N2 (seasonal), avian influenza A (H5N1, H5N2, H6N2, H7N2, H7N3, H2N2, H7N9, and H9N2), influenza B (representing strains from both Victoria and Yamagata lineages), and respiratory syncytial virus subgroups A and B (RSV A and RSV B) at levels near the analytical LoD. A total of 53 strains comprised of 48 influenza viruses (35 influenza A and 13 influenza B) and 5 RSV strains were tested in this study with the Xpert Xpress Flu/RSV Assay. Three replicates were tested for each strain. All Flu and RSV strains tested positive in all three replicates, except for one Flu A H1N1 strain (A/New Jersey/8/76), which tested positive in 2 of 3 replicates at 0.1 TCID₅₀/mL.

Results are shown in Table 8-9.

Further *in silico* analysis was conducted to determine the predicted cross reactivity of additional influenza A 2009 H1N1-like strains. The results showed 100% sequence homology for all primer target nucleotide sequences analyzed.

CONFIDENTIAL

Table 8-9 Analytical Reactivity (Inclusivity) of the Xpert Xpress Flu/RSV Assay

Virus	Strain	Target	Result		
			Flu A	Flu B	RSV
No Template Control		n/a	NEG	NEG	NEG
Influenza A H1N1 (pre-2009)	A/swine/Iowa/15/30	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/WS/33	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/PR/8/34	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Mal/302/54	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Denver/1/57	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/New Jersey/8/76	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/New Caledonia/20/1999	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/New York/55/2004	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Soloman Island/3/2006	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Taiwan/42/06	0.1 TCID ₅₀ /mL	POS	NEG	NEG
A/Brisbane/59/2007	0.1 TCID ₅₀ /mL	POS	NEG	NEG	
Influenza A H1N1 (pdm2009)	A/swine/NY/02/2009	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Colorado/14/2012	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Washington/24/2012	0.1 TCID ₅₀ /mL	POS	NEG	NEG
	A/Aichi/2/68	2.0 TCID ₅₀ /mL	POS	NEG	NEG

CONFIDENTIAL

Virus	Strain	Target	Result			
			Flu A	Flu B	RSV	
Influenza A H3N2 (Seasonal)	A/HongKong/8/68	2.0 TCID ₅₀ /mL	POS	NEG	NEG	
	A/Port Chalmers/1/73	2.0 TCID ₅₀ /mL	POS	NEG	NEG	
	A/Hawaii/15/2001	2.0 TCID ₅₀ /mL	POS	NEG	NEG	
	A/Wisconsin/67/05	2.0 TCID ₅₀ /mL	POS	NEG	NEG	
	Influenza A H3N2 (Seasonal)	A/Brisbane/10/2007	2.0 TCID ₅₀ /mL	POS	NEG	NEG
		A/Minnesota/11/2010 (H3N2)v	2.0 TCID ₅₀ /mL	POS	NEG	NEG
		A/Indiana/08/2011 (H3N2)v	2.0 TCID ₅₀ /mL	POS	NEG	NEG
		A/Texas/50/2012	2.0 TCID ₅₀ /mL	POS	NEG	NEG
		Avian influenza A	A/duck/Hunan/795/2002 (H5N1)	≤ 1pg/μL ^a	POS	NEG
A/chicken/Hubei/327/2004 (H5N1)	≤ 1pg/μL ^a		POS	NEG	NEG	
A/Anhui/01/2005 (H5N1)	≤ 1pg/μL ^a		POS	NEG	NEG	
A/Japanese white eye/HongKong/ 1038/2006	≤ 1pg/μL ^a		POS	NEG	NEG	
A/mallard/WI/34/75 (H5N2)	≤ 1pg/μL ^a		POS	NEG	NEG	
A/chicken/CA431/00 (H6N2)	≤ 1pg/μL ^a		POS	NEG	NEG	
A/duck/LTC-182743/1943 (H7N2)	≤ 1pg/μL ^a		POS	NEG	NEG	
A/chicken/NJ/15086-3/94 (H7N3)	≤ 1pg/μL ^a		POS	NEG	NEG	

CONFIDENTIAL

Virus	Strain	Target	Result		
			Flu A	Flu B	RSV
	A/Anhui/1/2013 (H7N9)	N/A _b	POS	NEG	NEG
	A/Shanghai/1/2013 (H7N9)	N/A _b	POS	NEG	NEG
	A/chicken/Korea/38349-p96323/1996 (H9N2)	$\leq 1\text{pg}/\mu\text{L}^{\text{a}}$	POS	NEG	NEG
	A/Mallard/NY/6750/78 (H2N2)	$\leq 1\text{pg}/\mu\text{L}^{\text{a}}$	POS	NEG	NEG
Influenza B	B/Lee/40	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Allen/45	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/GL/1739/54	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Maryland/1/59	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Panama/45/90 ^c	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Florida/07/2004 ^d	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Florida/02/06 ^c	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Florida/04/06 ^d	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Hong Kong/5/72	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Wisconsin/01/2010 ^d	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Malaysia/2506/04 ^c	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Taiwan/2/62	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	B/Brisbane/60/2008 ^c	1.0 TCID ₅₀ /mL	NEG	POS	NEG
	RSV-A/NY (Clinical unknown)	3.0 TCID ₅₀ /mL	NEG	NEG	POS

CONFIDENTIAL

Virus	Strai	Target	Result		
			Flu A	Flu B	RSV
RSV A	RSV-A/WI/629-8-2/2007	3.0 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-A/WI/629-11-1/2008	3.0 TCID ₅₀ /mL	NEG	NEG	POS
RSV B	RSV-B/WV14617/85	7.0 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-B/CH93(18)-18	7.0 TCID ₅₀ /mL	NEG	NEG	POS

- a. Purified viral RNA in simulated background matrix was used for avian influenza A viruses due to biosafety regulations.
- b. Inactivated avian influenza A (H7N9) viruses without viral titer was diluted 100,000 fold in simulated background matrix and tested due to biosafety regulations.
- c. Known Victoria lineage.
- d. Known Yamagata lineage.

Potentially Interfering Substances

In a non-clinical study, potentially interfering substances that may be present in the nasopharynx were evaluated directly relative to the performance of the Xpert Xpress Flu/RSV Assay. Potentially interfering substances in the nasopharynx may include, but are not limited to: blood, nasal secretions or mucus, and nasal and throat medications used to relieve congestion, nasal dryness, irritation, or asthma and allergy symptoms, as well as antibiotics and antivirals. Negative samples (n = 8) were tested per each substance to determine the effect on the performance of the sample processing control (SPC). Positive samples (n = 8) were tested per substance with six influenza (four influenza A and two influenza B) and four RSV (two RSV A and two RSV B) strains spiked at 3X the analytical LoD determined for each strain. All results were compared to positive and negative simulated nasal matrix controls. The simulated nasal matrix consisted of 2.5% (w/v) porcine mucin, 1% (v/v) human whole blood in 0.85% sodium chloride (NaCl) formulated in 1x PBS solution with 15% glycerol, which was then diluted 1:5 in UTM. The evaluated substances are listed in Table 8-10 with active ingredients and concentrations tested shown. None of the substances caused interference of the assay at the concentrations tested in this study. All positive and negative replicates were identified correctly using the Xpert Xpress Flu/RSV Assay.

CONFIDENTIAL

Table 8-10 Potentially Interfering Substances in the Xpert Xpress Flu/RSV Assay

Substance/Class	Description/Active Ingredient	Concentration Tested
Control	Simulated nasal matrix	100% (v/v)
Beta-adrenergic bronchodilator	Albuterol Sulfate	0.83 mg/mL (equivalent to 1 dose per day)
Blood	Blood (Human)	2% (v/v)
BD™ Universal Viral Transport System	Transport Media	100% (v/v)
Remel M4®	Transport Media	100% (v/v)
Remel M4RT®	Transport Media	100% (v/v)
Remel M5®	Transport Media	100% (v/v)
Remel M6®	Transport Media	100% (v/v)
Throat lozenges, oral anesthetic and analgesic	Benzocaine, Menthol	1.7 mg/mL
Mucin	Purified Mucin protein (Bovine or porcine submaxillary gland)	2.5% (w/v)
Antibiotic, nasal ointment	Mupirocin	10 mg/mL
Saline Nasal Spray	Sodium Chloride (0.65%)	15% (v/v)
Anefrin Nasal Spray	Oxymetazoline, 0.05%	15% (v/v)
PHNY Nasal Drops	Phenylephrine, 0.5%	15% (v/v)
Tamiflu Anti-viral drugs	Zanamivir	7.5 mg/mL
Antibacterial, systemic	Tobramycin	4 µg/mL
Zicam Nasal Gel	Luffa operculata, Galphimia glauca, Histaminum hydrochloricum Sulfur	15% (w/v)
Nasal corticosteroid	Fluticasone Propionate	5 µg/mL

Carry-Over Contamination

A study was conducted to demonstrate that single-use, self-contained GeneXpert cartridges prevent carry-over contamination of negative samples when followed by very high positive samples in the same GeneXpert module. The study consisted of a negative sample processed in the same GeneXpert module immediately followed by a very high influenza A sample (A/Victoria/361/2011, 2×10^7 TCID₅₀/mL) or a very high RSV A sample (A/Long/MD/26, 1×10^4 TCID₅₀/mL) spiked into a simulated nasal matrix. This testing scheme was repeated 20 times on two GeneXpert modules for a total of 82 runs resulting in 40 positive and 42 negative specimens for each virus type. All 40 positive samples were correctly reported as Flu A POSITIVE; Flu B NEGATIVE; RSV NEGATIVE or Flu A NEGATIVE; Flu B NEGATIVE; RSV POSITIVE. All 42 negative samples were correctly reported as Flu A NEGATIVE; Flu B NEGATIVE; RSV NEGATIVE.

Fresh vs. Frozen Sample Equivalency Study

Fresh and frozen specimen equivalency in the Xpert Xpress Flu/RSV Assay was evaluated by testing individual influenza and RSV strains at three different concentrations representing low positives (2X LoD), moderate positives (5X LoD), and high positives (10X LoD) in pooled negative NP swab clinical matrix. Negative samples consisted of pooled negative NP swab clinical matrix only. Fresh and frozen specimen equivalency was determined using one seasonal Flu A H3N2 strain (A/Victoria/361/2011), one Flu B strain (B/Mass/2/2012), one RSV A strain (RSV-A/2/Australia/61), and one RSV B strain (RSV-B/Wash/18537/62). Replicates of 20 were tested for each specimen type and concentration. All positive and negative specimens were tested fresh, after one freeze-thaw cycle, and after two freeze-thaw cycles. There was no difference in the performance of the Xpert Xpress Flu/RSV Assay between fresh virus dilutions and two sequential freeze thaw

CONFIDENTIAL

cycles for positive and negative samples. All positive and negative replicates were correctly identified using the Xpert Xpress Flu/RSV Assay.

Competitive Interference Study

Competitive interference of the assay caused by the presence of two targets in the Xpert Xpress Flu/RSV Assay was evaluated by testing individual influenza and RSV strains near the LoD in the presence of different influenza or RSV strains at a higher concentration in a simulated nasal matrix. The concentration of each strain at LoD ranged from 0.45 TCID₅₀/mL to 1.6 TCID₅₀/mL and the concentration of the competitive strains ranged from 10¹ TCID₅₀/mL to 10⁴ TCID₅₀/mL. Analytical competitive interference was assessed using one (1) seasonal Flu A H3 strain (H3/Victoria/361/2011), one (1) Flu B strain (B/Mass/2/2012), one (1) RSV A strain (RSV-A/2/Australia/61), and one (1) RSV B strain (RSV-B/Wash/18537/62). Replicates of 20 were tested for each target strain and each competitive strain combination. The normal binomial distribution with 20 replicate samples at LoD is between 17 and 20 positive results based on the binomial distribution with N=20, p=.95 (X~Bin(20,0.95)). Therefore, sets of 20 with 16 or less positives would be rare and an indication of a competitive inhibitory effect due to high levels of a competing analyte.

- With Flu A/Victoria/361/2011 at a concentration of 0.8 TCID₅₀/mL no competitive inhibitory effects were observed in the presence of 1x10³ TCID₅₀/mL of Flu B/Mass/2/2012; 1x10³ TCID₅₀/mL of RSV-A/2/Australia/6; or 1x10⁴ TCID₅₀/mL of RSV-B/Wash/18537/62.
- With Flu B/Mass/2/2012 at a concentration of 0.45 TCID₅₀/mL competitive inhibitory effects were observed in the presence of 1x10³ TCID₅₀/mL of Flu A/Victoria/361/2011. No competitive inhibitory effects were observed in the presence of 1x10² TCID₅₀/mL of Flu A/Victoria/361/2011; 1x10³ TCID₅₀/mL of RSV-A/2/Australia/6; or 1x10³ TCID₅₀/mL of RSV-B/Wash/18537/62.
- With RSV-A/2/Australia/6 at a concentration of 1.1 TCID₅₀/mL competitive inhibitory effects were observed in the presence of 1x10³ TCID₅₀/mL of Flu

CONFIDENTIAL

A/Victoria/361/2011. No competitive inhibitory effects were observed in the presence of 1×10^2 TCID₅₀/mL of Flu A/Victoria/361/2011; or 1×10^3 TCID₅₀/mL of Flu B/Mass/2/2012.

- With RSV-B/Wash/18537/62 at a concentration of 0.9 TCID₅₀/mL competitive inhibitory effects were observed in the presence of 1×10^2 TCID₅₀/mL of Flu A/Victoria/361/2011 or 1×10^3 TCID₅₀/mL of Flu B/Mass/2/2012. No competitive inhibitory effects were observed in the presence of 10 TCID₅₀/mL of Flu A/Victoria/361/2011; or 1×10^2 TCID₅₀/mL of Flu B/Mass/2/2012. When the concentration of RSV-B/Wash/18537/62 was increased to 1.6 TCID₅₀/mL, no competitive inhibitory effects were observed in the presence of 1×10^2 TCID₅₀/mL of Flu A/Victoria/361/2011; or 1×10^3 TCID₅₀/mL of Flu B/Mass/2/2012.
- Under the conditions of this study, internal competitive inhibitory effects were observed on the targets (Flu A, Flu B, and RSV) in the presence of two targets for the Xpert Xpress Flu/RSV Assay. The competitive inhibitory effect on the Xpert Xpress Flu/RSV targets is addressed in the Limitations section of the package insert.

Clinical Studies

Clinical Comparison Study

Performance characteristics of the Xpert Xpress Flu/RSV Assay were evaluated at eleven institutions in the U.S. during the 2015-2016 influenza season. Due to the low prevalence of influenza viruses and the difficulty in obtaining fresh influenza and RSV-positive specimens, the specimen population for this study was supplemented with consecutively collected, frozen specimens.

Specimens were collected from the following:

- Individuals exhibiting signs and symptoms of respiratory infection who provided informed consent for the collection of a NP swab specimen.

CONFIDENTIAL

- Individuals with signs and symptoms of respiratory infection and whose routine care called for collection of NP swab specimens for influenza and/or RSV testing. For eligible subjects, aliquots of leftover specimens were obtained for testing with the Xpert Xpress Flu/RSV Assay and reference testing, and patient management continued at the site per their standard practice.

The Xpert Xpress Flu/RSV Assay performance was compared to FDA-cleared molecular comparator assay. Bi-directional sequencing was performed on specimens where the Xpert Xpress Flu/RSV Assay and the comparator assay were discrepant, and is provided for informational purposes only.

Overall Results – NP Swab Specimens

A total of 2065 NP swab specimens were tested for influenza A, influenza B and RSV by the Xpert Xpress Flu/RSV Assay and the comparator assay. Of the 2065 NP swab specimens, 1142 were fresh, prospectively collected and 923 were consecutively collected, frozen specimens.

For the fresh, prospectively collected NP swab specimens, the Xpert Xpress Flu/RSV Assay demonstrated a PPA and NPA of 94.6% and 99.3%, detection of influenza A; 100% and 99.1% for influenza B, respectively; and 100% and 99.6%, for RSV, respectively, relative to the comparator assay (Table 8-11).

For the consecutively collected, frozen NP swab specimens, the Xpert Xpress Flu/RSV Assay demonstrated a PPA and NPA of 100% and 97.2% for the detection of influenza A, respectively; 100% and 98.2% for influenza B, respectively; and 97.9% and 97.7% for RSV, respectively, relative to the comparator assay (Table 8-11).

For the combined dataset, relative to the comparator assay, the Xpert Xpress Flu/RSV Assay demonstrated a PPA and NPA of 98.1% and 98.4% for the detection of influenza A, respectively; 100% and 98.7% for influenza B respectively; and 98.5% and 98.8% for RSV, respectively (Table 8-11).

CONFIDENTIAL

Table 8-11 Xpert Xpress Flu/RSV Assay Performance on NP Swab Specimens

Specimen Type	Target	N	TP	FP	TN	FN	PPA (95% CI)	NPA (95% CI)
Fresh, prospectively collected	Flu A	1142	35	8 ^a	1097	2 ^b	94.6% (CI:82.3-98.5)	99.3% (CI:98.6-99.6)
	Flu B	1142	42	10 ^c	1090	0	100.0% (CI:91.6-100.0)	99.1% (CI:98.3-99.5)
	RSV	1142	17	5 ^d	1120	0	100.0% (CI:81.6-100.0)	99.6% (CI:99.0-99.8)
Frozen, consecutively collected	Flu A	923	69	24 ^e	830	0	100.0% (CI:94.7-100.0)	97.2% (CI:95.9-98.1)
	Flu B	923	36	16 ^f	871	0	100.0% (CI:90.4-100.0)	98.2% (CI:97.1-98.9)
	RSV	923	47	20 ^g	855	1 ^h	97.9% (CI:89.1-99.6)	97.7% (CI:96.5-98.5)
Combined ¹	Flu A	2065	104	32 ⁱ	1927	2 ^b	98.1% (CI:93.4-99.5)	98.4% (CI:97.7-98.8)
	Flu B	2065	78	26 ^j	1961	0	100.0% (CI:95.3-100.0)	98.7% (CI:98.1-99.1)
	RSV	2065	64	25 ^k	1975	1 ^h	98.5% (CI:91.8-99.7)	98.8% (CI:98.2-99.2)

a. Testing results by sequencing: 3 of 8 were Flu A Positive; 4 of 8 were Flu A Negative; 1 of 8 insufficient specimen for sequencing.
b. Testing results by sequencing: 2 of 2 were Flu A Negative.
c. Testing results by sequencing: 6 of 10 were Flu B Positive; 3 of 10 were Flu B Negative; 1 of 10 insufficient specimen for sequencing.
d. Testing results by sequencing: 0 of 5 were RSV Positive; 3 of 5 were RSV Negative; 2 of 5 insufficient specimen for sequencing. e. Testing results by sequencing: 8 of 24 were Flu A Positive; 11 of 24 were Flu A Negative; 5 of 24 insufficient specimen for sequencing.
f. Testing results by sequencing: 7 of 16 were Flu B Positive; 3 of 16 were Flu B Negative; 6 of 16 insufficient specimen for sequencing.
g. Testing results by sequencing: 3 of 20 were RSV Positive; 8 of 20 were RSV Negative; 9 of 20 insufficient specimen for sequencing.
h. Testing results by sequencing: 1 of 1 was RSV Negative.
i. Testing results by sequencing: 11 of 32 were Flu A Positive; 15 of 32 were Flu A Negative; 6 of 32 insufficient specimen for sequencing.
j. Testing results by sequencing: 13 of 26 were Flu B Positive; 6 of 26 were Flu B Negative; 7 of 26 insufficient specimen for sequencing.
k. Testing results by sequencing: 3 of 25 were RSV Positive; 11 of 25 were RSV Negative; 11 of 25 insufficient specimen for sequencing
l. Nine specimens (8 Flu A FP; 9 Flu B FP; 7 RSV FP) were positive for all three targets.

Of the Xpert Xpress Flu/RSV Assay runs performed with eligible specimens, 98.4% (2038/2071) of these specimens were successful on the first attempt. The remaining 33 gave indeterminate results on the first attempt (20 **ERROR**, 10 **INVALID**, and 3 **NO RESULT**). The initial indeterminate rate was 1.59% (33/2071) with the 95% CI 1.14-2.23%. Thirty of the 33 indeterminate cases were retested, of which 27 yielded valid results upon repeat testing; three specimens were not retested. The overall rate of assay success was 99.7% (2065/2071). The overall indeterminate rate after retesting was 0.3% (6/2071) with 95% CI 0.13- 0.63%.

In addition, there were 102 pre-selected frozen NP swab specimens tested. The results of this testing were analyzed separately and are as follows: the Xpert Xpress Flu/RSV Assay demonstrated a PPA and NPA of 100% and 95.8%, for influenza A, respectively; 100% and 94.5% for influenza B, respectively; and 100% and 95.7%, for RSV, respectively, relative to the comparator assay.

Reproducibility Study

Reproducibility was established in a multi-center, blinded study using a seven-member specimen panel consisting of a negative control and two each of simulated nasal matrix spiked with influenza A, influenza B or RSV at 1X (low pos) and 2-3X (mod pos) the respective LODs. Testing was performed at three sites (one internal, two external) using the GeneXpert Dx system, the Infinity-48 system, and the Infinity-80 system. Two operators at each site tested one panel in duplicate two times per day (equivalent to four replicates per day) over six, not necessarily consecutive days. Three lots of Xpert Xpress Flu/RSV cartridges were used, with each lot representing approximately two days of testing. Results are summarized in Table 8-12.

CONFIDENTIAL

Table 8-12 Summary of Reproducibility Results

Sample ID	Site 1/Infinity-80			Site 2/DX			Site 3/Infinity-48			% Total Agreement by Sample ^a
	Op 1	Op 2	Site	Op 1	Op 2	Site	Op 1	Op 2	Site	
Negative	100% (24/24)	100% (24/24)	100% (48/48)	100% (24/24)	100% (24/24)	100% (48/48)	100% (24/24)	100% (24/24)	100% (48/48)	100% (144/144)
Flu A- Low Pos	87.5% (21/24)	95.8% (23/24)	91.7% (44/48)	95.7% (22/23)	91.7% (22/24)	93.6% (44/47)	100% (24/24)	91.7% (22/24)	95.8% (46/48)	93.7% (134/143) ^b
Flu A- Mod Pos	100% (24/24)	100% (24/24)	100% (48/48)	100% (23/23)	100% (23/23)	100% (46/46)	100% (24/24)	100% (24/24)	100% (48/48)	100% (142/142) ^b
Flu B- Low Pos	95.8% (23/24)	95.8% (23/24)	95.8% (46/48)	95.8% (23/24)	95.8% (23/24)	95.8% (46/48)	95.8% (23/24)	91.7% (22/24)	93.8% (45/48)	95.1% (137/144)
Flu B- Mod Pos	95.8% (23/24)	100% (24/24)	97.9% (47/48)	100% (24/24)	100% (24/24)	100% (48/48)	100% (24/24)	95.8% (23/24)	97.9% (47/48)	98.6% (142/144) ^c
RSV- Low Pos	91.7% (22/24)	87.5% (21/24)	89.6% (43/48)	100% (23/23)	100% (24/24)	100% (47/47)	91.7% (22/24)	95.8% (23/24)	93.8% (45/48)	94.4% (135/143) ^b
RSV- Mod Pos	100% (24/24)	100% (24/24)	100% (48/48)	100% (23/23)	100% (24/24)	100% (47/47)	100% (24/24)	100% (24/24)	100% (48/48)	100% (143/143) ^b

- a. Agreement calculated based on expected result: Negative for Negative (targeted positivity: 0%); Positive for Low Pos (targeted positivity: 95%) and Mod Pos (targeted positivity: 100%) samples.
- b. Fives samples 2x indeterminate [Flu A Low Pos (1); Flu A Mod Pos (2); RSV Low Pos (1); RSV Mod Pos (1)]
- c. Two Flu B Mod Pos samples were positive for all three targets

The reproducibility of the Xpert Xpress Flu/RSV Assay was also evaluated in terms of the fluorescence signal expressed in Ct values for each target detected. The mean, standard deviation (SD), and coefficient of variation (CV) between-sites, between- days, between-lots and between-operators for each panel member are presented in Table 8-13.

CONFIDENTIAL

Table 8-13 Summary of Reproducibility Data

Sample	Assay Channel (Analyte)	N ^a	Mean Ct	Between-Site		Between-Lot		Between-Day		Between-Operator		Within-Assay		Total	
				SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)
Negative	SPC	144	32.3	0	0	0.7	2.1	0	0	0.2	0.5	0.6	1.8	0.9	2.8
Flu A-Low Pos	FluA1	134	35.3	0	0	0.4	1.1	0.6	1.8	0.1	0.4	0.9	2.5	1.2	3.3
Flu A-Mod Pos	FluA1	142	33.1	0	0	0.1	0.4	0.1	0.4	0	0	0.6	1.9	0.7	2.0
Flu B-Low Pos	FluB	137	34.6	0	0	0	0	0.4	1.3	0	0	1.4	4.1	1.5	4.3
Flu B-Mod Pos	FluB	144	32.2	0.2	0.5	0.2	0.7	0	0	0.2	0.7	1.0	3.1	1.1	3.3
RSV-Low Pos	RSV	135	36.5	0	0	0.6	1.7	0	0	0.5	1.3	0.9	2.6	1.2	3.3
RSV-Mod Pos	RSV	143	33.5	0.2	0.7	0.1	0.4	0	0	0	0	1.5	4.6	1.6	4.6

a. Results with non-zero Ct values of 144.

Conclusions

The results of the nonclinical analytical and clinical performance studies summarized above demonstrate that the Xpert Xpress Flu/RSV Assay is substantially equivalent to the predicate device.